

# Bacterial and Fungal Contamination of Air conditioners filters and Carpets

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## Abstract— Background:

**Objective:** To study the level of colonization by bacteria, and fungi/molds in air conditioners and carpets used in homes, offices, university classrooms, prayer room and laboratories.

**Methods:** A total of 25 different settled dust samples from air conditioners, and carpet vacuum cleaners were investigated in this study. One gram of each sample was emulsified into 9.0mL sterile normal saline. 10-fold dilutions were made in normal saline and 100  $\mu$ L of each suspension was inoculated onto 2 plates of Sabouraud Dextrose Agar incubated at 25 °C for 48-72hrs, while 50  $\mu$ L was inoculated onto MacConkey and Blood agar plates which were incubated at 37 °C for 48hrs.

**Results:** of the settled dust samples examined, 72 % were found to harbor molds, bacteria and yeasts. Of the total bacteria isolated, *Bacillus* spp. comprised 60.3%, *S. epidermidis* (20.7%), *S. aureus* (17.2%), Gram negative bacilli (1.7%). On the other hand, *Aspergillus* spp. made up 26.9%, of the total mold isolates, followed by *Penicillium* spp. (20.9%), Yeast (13.4%), *Rhizopus* spp. (7.5%), *Zygomycetes* spp. (3%), *Aspergillus niger* (3%), *Lichtheimia corymbifera* (3%), *Microsporum* spp. (1.5%) and Unidentified mold in 20.8%. The mean bacterial concentration found was  $1.33 \times 10^5$  Cfu/g of dust.

**Conclusion:** The presence of potentially pathogenic bacteria and molds in the dust samples of the air conditioner filters, and home carpets indicated that our environment is full of microorganism that may affect people's health and expose them to different diseases, this mandates regular cleaning and safe removal of dust from A/C filters and carpets.

**Keywords**—air conditioner filters, carpets, bacteria, Fungi.

## I. INTRODUCTION

Our planet is surrounded by air and dust where variety of microorganisms are found. Aerobiology is the study of the airborne microorganisms and their effect on human health and the environment. As a result of increasing awareness of the variety of health problems caused by airborne microorganisms [1] research in this area has been increasing recently. Airborne contaminants are also known as bioaerosols and include bacteria, fungi, viruses and

pollen. These may be present in the air as solid (dust) or as liquid (condensation and water). An aerosol is a two-phase system of gaseous phase (air) and particulate matter (dust, pathogens), thus making an important bacterial vehicle. The micro organisms that can be found in bioaerosols can be found attached to dust particles or may survive as freefloating particles surrounded by a coating of dried organic or inorganic material and due to a lack of nutrients they cannot multiply in bioaerosols but they can travel in the air for great distances [2, 3].

The developmental process of using air conditioners and carpets by humans in houses, class rooms, mosques and other places though resulted in the help of easing life for humans during times when weather is harsh, however, their use unfortunately brought undesirable effects in that they became a good source and or media for bacterial and fungal/mold contamination [4]. The improperly maintained air conditioning systems and exposure to these microbial fragments and metabolites can be a source of wide variety of illnesses ranging from allergy symptoms to dangerous conditions such as respiratory, dermatological infections, epidemics and food pollution [5]. In particular, individuals with poor health or immune compromised patients are at a greater risk to suffer from exposure to these microorganisms that could be present in this environment [6]. Recent research findings have shown that certain vacuum cleaners unfortunately can make things worse, in that they have the ability to spit fine dust back into the air, where they can spread any kind of infections [7] in the surrounding environment, which leads to the belief that microorganisms are linked to poor indoor air quality [8].

It has been estimated in Northern Europe and Canada that 20-40% of homes have mold contamination [9]. This number in tropical and subtropical countries is estimated to be much higher [10, 11]. Whereas, in United States, as many as 47% of homes have mold contamination [12]. Various health effects have been related to mold exposure [13]. A case control study conducted in Europe suggested a relation between increased symptoms of asthmatic patients and increased mold and moisture problems in homes [14]. Jo and Lee (2008), [15] reported that automobile and household air conditioning can discharge up to 2,500 colony forming units per meter

cubed (CFU/m<sup>3</sup>) of bacteria, as well as 1,000 CFU/m<sup>3</sup> of fungi at ambient levels during initial startup. Conspicuous growth (up to 10<sup>6</sup>CFU/cm<sup>2</sup>) of bacteria is reported from air handling cooling coils.

Ali et.al., (2014)[16] have investigated the presence of bacteria, fungi and mold in 50 dust samples from carpets of 50 mosques in Elkhomes city in Libya, and found that 12% were positive for *Escherichia coli*, 60% for *Staphylococcus* spp. and 66% for *Klebsiella* spp. Similarly, Rahouma et.al., (2010) [17] have collected 57 dust samples from carpets of mosques in Tripoli city in Libya, and reported the presence of *E. coli* in 16 samples, *Staphylococcus aureus* in 12 sample, *Salmonella* spp. in 2 samples and *Aeromonas* spp. in one sample.

The aim of the present study is to determine the presence of bacteria and fungi in settled dust samples collected from home carpets and air conditioners filters in different places in Sakaka city, Aljouf, Saudi Arabia.

## II. MATERIALS AND METHODS

### Location of study:

Sakaka is located in the northeast of Saudi Arabia. The geographical location is between longitudes 37/42 east and latitudes 29/32 north, and it rises from the sea at 580 feet with an estimated area of about 107.794 km<sup>2</sup>, which is equivalent to 4.9% of the total area of the Saudi Arabia. Climate fluctuations are evident with the average maximum temperature of 42 °C in the summer, and the average winter temperature of 8.5 °C. The month of July is the hottest months of the year, while temperatures drop in the month of January with an average rainfall of approximately 200 mm. A north-west wind is reported in the area during the month of April, whereas, an eastern-western wind in the rest of other months. The average speed of wind is about 5.7 miles per hour.

### Sample collection:

A total of 25 settled dust samples were collected from carpet's vacuum cleaners (*number=5*) and filters of air conditioners (*number=20*). Sampling was performed during the period Dec 2015 to Feb 2016. The carpet dust samples were collected using a 2,000 W and 1400 W household vacuum cleaners; while samples from filters of air conditioners were collected from Window type A/C, Split A/C and Tower A/C. Settled dust was carefully removed from the filters and collected onto a clean paper and transferred into sterile petri dishes and transported to the microbiology laboratory at the college of Medicine, Aljouf University, and processed within 1-10 hours of collection.

### Bacteriology

After collection, the contents (i.e. dust, settled material, hair, etc.) of each sample was thoroughly mixed together

and 1.0 g of each sample was weighed and emulsified into 9.0 ml of sterile normal saline, and vortexed for 2 minutes to ensure that all the contents were mixed well and to disperse any microorganisms that may have been trapped into the dust. A ten-fold dilution of each sample (10<sup>1</sup> - 10<sup>2</sup>) was performed using sterile normal saline, and 100 µL of each dilution was transferred and spread onto two *Sabouraud Dextrose Agar* plates (SDA) (Oxoid LTD., Basingstoke, Hampshire-England) using a glass spreader. One plate each of *MacConkey agar* (Oxoid LTD., Basingstoke, Hampshire-England) and *Blood Agar* (Oxoid LTD., Basingstoke, Hampshire-England) were streaked using 50 µl of each sample. All plates were incubated at 37°C in air for 24 hours, except for SDA plates which were incubated at 25°C for 48-72 hrs. [18]

After incubation, any visibly distinguishable bacterial or fungal colonies were identified and the number on each plate was counted. The initial observations about the shape, color, size and other visual properties of each isolate were also recorded. A representative sample colony of each visually differentiable bacteria, fungi or mold was selected using a sterile inoculating loop, and was transferred by streaking onto a fresh plate of SDA, Blood agar or MacConkey agar, and incubated as previously described. All bacterial isolates were Gram stained and examined under the microscope using the X1000 lens. [19]. Any suspected yeast's or molds were examined using a drop of lacto phenol cotton blue stain under the X400 lens.

## III. IDENTIFICATION OF BACTERIA AND FUNGAI

### Biochemical tests:

In order to identify the isolated bacteria some biochemical tests were used including:

**Catalase test:** To test for the presence of catalase, isolated colonies were selected and transferred via a sterile inoculating plastic loop to a clean slide containing one drop of hydrogen peroxide. If the bacteria contained catalase, hydrogen peroxide was converted to water and oxygen gas, causing bubbles to appear on the slide. Catalase negative bacteria produce no reaction [20].

**Coagulase test:** Is an enzyme-like protein and causes plasma to clot by converting fibrinogen to fibrin; using a plastic pipette aseptically 0.5 ml of 1:3 diluted human plasma was transferred into a test tube, the isolated colonies of the bacteria to be tested were emulsified into the plasma and incubated at 37°C for 3 - 4 hours. Any clot formation was taken as a positive result, however, if no clot was observed by the end of 4 hours, then the test was continued with incubated at 37°C and a final observation was taken at 24 hours [21].

**Results:**

Of the 25 samples examined in this study, 20 (80%) were made of settled dust collected from different types of A/C filters; while the remaining 20% represented dust from carpet vacuum cleaners (Figure 1). The amount of settled dust collected was greater in the vacuum cleaners than window type air conditioners. However, moderate amounts or very little settled dust was evident on the split or the tower type A/C.

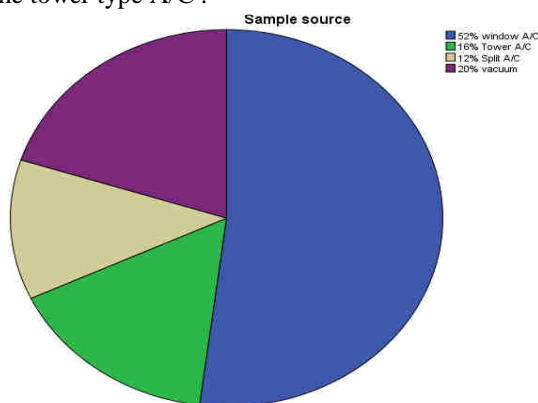


Fig.1: Percentages and sources of the settled dust samples.

As shown in Table 1, of the total 58 bacterial isolates 60% were identified as *Bacillus* species, 20.7% as *S. epidermidis*; 17% *S. aureus* and only 1.7% was identified as Gram negative bacilli's. On the other hand, a total of 67 isolates were recovered from both the A/C filters and carpet samples. Of the total mold/yeast isolates 26.9% were identified as *Aspergillus* spp. followed by *Penicillium* spp. (20.9%), *Rhizopus* spp. (7.4%) and yeast in 13.4%. While other molds were found in smaller percentages as shown in Table 2. Using the cotton blue technique 20.8% of the total mold isolates could not be fully identified.

Table.1: Bacteria isolated from A/C filters and carpets

Microorganisms	Frequency (%)
<i>Bacillus</i> spp.	35 (60.3%)
<i>S. epidermidis</i>	12 (20.7%)
<i>S. aureus</i>	10 (17.2%)
Gram negative bacilli	1 (1.7%)
Total isolates	58 (100%)

Table.2: Molds and Yeast isolated from A/C filters and carpets.

Microorganisms	Frequency (%)
<i>Aspergillus</i> spp.	18 (26.9%)
<i>Penicillium</i> spp.	14 (20.9%)
Yeast	9 (13.4%)

<i>Rhizopus</i> spp.	5 (7.5%)
<i>Zygomycetes</i> spp.	2 (3%)
<i>Aspergillus niger</i>	2 (3%)
<i>Lichtheimiacorymbifera</i>	2 (3%)
<i>Microsporum</i> spp.	1 (1.5%)
Unidentified mold	14 (20.8%)
Total isolates	67 (100%)

Table.3: Total viable count of the bacterial isolates on McConkey and Blood agar plates.

Total viable count	Number of Samples	Minimum Viable count (cfu/g)	Maximum Viable count (cfu/g)	Mean Viable count (cfu/g)	Standard Deviation
McConkey agar	25	2500	420000	132804	128190
Blood agar	25	11000	520000	133400	129202

The concentration of the viable bacterial count on McConkey agar ranged from  $2.5 \times 10^3$  to  $4.2 \times 10^5$  Cfug of settled dust (mean  $1.3 \times 10^5$  Cfug). While the use of blood agar yielded a growth concentration that ranged from  $1.1 \times 10^4$  to  $5.2 \times 10^5$  Cfug (mean  $1.33 \times 10^5$  Cfug).

#### IV. DISCUSSION

Microorganisms such as bacteria, molds and yeast are common contaminants in our environment. The inappropriately maintained air conditioning systems and exposure to metabolites can be a source of wide variety of diseases such as allergy, respiratory, dermatological infections, epidemics and food contamination [4]. In view of the climate changes in the northern part of Saudi Arabia, almost all inhabitants have adopted to the use of A/C at homes, work place, prayer places and in their automobiles. As reported by Abdel Hameed and Farag in 1999[22], most of the people in arid areas tend to spend over 90% of their lives indoors: in houses, offices, and schools where they are exposed to some indoor environmental factors (bioaerosols) that influence their health and physical condition. Fungi reproduce through the production of microscopic spores, many of which are dispersed bywind, rainfall, and physical disturbance. Biological contamination of indoor air is mostly caused by bacteria, molds and yeast. In addition to being

dangerous as pathogenic living cells they can also secrete some substances harmful for health.

As reported in the present study that almost all the dust samples collected from the A/C filters were colonized by bacteria, yeast and molds. This colonization by these microorganisms is partially explained by the fact that condensation of water and increase in relative humidity and temperature in the A/C favors fungal growth. As a common practice the A/C units examined in this study are usually switched on and off during the day, this mandates frequent cleaning to reduce the clogging of the filters by dust and to reduce the likely chance of fungal or bacterial growth [23, 24]. Similarly, home carpets were found to harbor bacteria, yeast and molds, a periodic shampooing and vacuum cleaning is also necessary in order to minimize the likely hood of microbial colonization [25]. Although, the common way to clean home carpets is by the use of vacuum cleaners, however, their use has been associated with the release of high concentrations of antigens through the mechanical disturbance of settled dust and release from the vacuum cleaner itself [26]. The results of the present investigation has shown that dust samples collected from carpets contained several types of bacteria including *S. aureus*, *S. epidermidis*, Gram negative bacilli, and other types of molds such as *Aspergillus* and *Penicillium* spp. and others which is in agreement with the findings by other researchers [27]. It has also been reported that aerosolization of carpet dust can be a source to spread other pathogenic bacteria such as *Salmonella* and *Clostridium botulinum* [28]. What make things worse is that the presence of such microorganisms in dust bags or the chambers of the vacuum cleaners can be extended over long periods of time [29].

The amount of dust present in the A/C and carpets varied considerably. Obviously, vacuum and A/C filters samples exhibited too much amount of dust in comparison with Tower A/C filters, where the least amount of dust was found. As shown in the results section the concentration of the different microorganisms obtained on both McConkey agar and blood agar plates didn't significantly differ as the mean number of viable colonies was comparable on both types of media. Of the bacteria isolated in the different samples of dust, *Bacillus* spp. Represented 60.3% of the total bacterial isolates followed by *S. epidermidis* (20.7%), *S. aureus* (17.2%) and GNB (1.7%). however, a study conducted in Libya by Ali et.al., (2014) [16] showed the presence of other microorganisms such as *E. coli* and *Klebsiella* spp. This can be explained by the fact that the source of the dust samples examined in both studies differed. The Libyan samples were collected from carpets at different mosques where large group of worshipers usually gather to

perform religious duties. Whereas, in the present study a smaller number of samples from home carpets were used. It also expected that a better cleaning and care is taking at homes which can reduce the amount of dust and therefore bacterial and fungal or mold presence. Other pathogenic bacteria were also isolated from Masjed (mosques) carpet samples. For instance Rahouma et.al., (2010) [17] reported the presence of *E. coli* in 16 samples, *Staphylococcus aureus* in 12 sample, *Salmonella* spp. in 2 samples and *Aeromonas* spp. in one sample. Our study has shown the presence of *S. aureus* in a number of samples; however, the predominant bacterial species present were *Bacillus* spp. Of the 25 samples examined in our study 18/25 (72%) of the samples yielded the growth of *Aspergillus* spp. which represented 26.9% of the total mold and fungal isolates. Similarly, *Penicillium* spp. were largely found making up 20.9%, and Yeast 13.4%. We were unable to identify 16.4% of the molds present using the colonial morphology and microscopical examination of these isolates. In a study conducted in Warsaw, Poland by Gołofit-Szymczak & Górny [30], they found almost similar results were *Aspergillus* spp. and *Penicillium* were the most common isolates they encountered. The presence of certain bacterial species such as *S. aureus* and *S. epidermidis* on carpets is highly suspected, because these bacteria are present in the nose and skin of people as part of their normal flora [31]. In view of the sources of the settled dust samples used in our study and others, it was not possible to make a direct comparison of results.

## V. CONCLUSIONS AND RECOMMENDATIONS

The presence of potentially pathogenic bacteria and molds or yeast in the dust samples of the examined air conditioner filters, and home carpets indicated that our environment is full of microorganism that may affect people's health and expose them to risk of different diseases, this mandates the regular cleaning and removal of dust from A/C filters and carpets.

The use of good quality vacuum cleaners is also recommended to decrease the chance of people being exposed to indoor allergen or microorganisms. It is likewise highly advised to keep cleaning of the air conditioners and vacuums regularly, and changing of the carpet and other furniture. Further studies are recommended to a follow up.

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